(FILE 'HCAPLUS, MEDLINE, BIOSIS, EMBASE, WPIDS, CONFSCI, SCISEARCH, JICST-EPLUS, JAPIO, CABA, AGRICOLA, FSTA, CROPU, CROPB' ENTERED AT 10:34:50 ON 27 MAR 2003)

Author(s)

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L12          118 SEA ABB=ON  PLU=ON  "BARBAS III C"?/AU
L13           41 SEA ABB=ON  PLU=ON  "STEGE J"?/AU
L14         2369 SEA ABB=ON  PLU=ON  "GUAN X"?/AU
L15         1078 SEA ABB=ON  PLU=ON  "BARBAS C"?/AU
L16           14 SEA ABB=ON  PLU=ON  (L12 OR L15) AND L13 AND L14
L17           15 SEA ABB=ON  PLU=ON  (L12 OR L15) AND (L13 OR L14)
L18           14 SEA ABB=ON  PLU=ON  L13 AND L14
L19           87 SEA ABB=ON  PLU=ON  (L12 OR L15 OR L13 OR L14) AND (ZFP?
                 OR (ZF OR (ZN OR ZINC)(W) FINGER)(W) PROTEIN)
L20           35 SEA ABB=ON  PLU=ON  L19 AND (PLANT OR MAIZE OR CORN OR
                 CARROT OR TOBACCO OR TOMATO OR POTATO OR BANANA OR
                 SOYABEAN OR SOYBEAN OR (SOY OR SOYA)(W) BEAN OR PEPPER
                 OR WHEAT OR RYE OR RICE OR SPINACH)
L21           36 SEA ABB=ON  PLU=ON  L16 OR L17 OR L18 OR L20
L22           13 DUP REM L21 (23 DUPLICATES REMOVED)
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L22 ANSWER 1 OF 13 HCAPLUS COPYRIGHT 2003 ACS

ACCESSION NUMBER: 2003:133983 HCAPLUS
DOCUMENT NUMBER: 138:182057
TITLE: Usage of **zinc finger proteins** and their fusions with effector domains to regulate gene expression and metabolic pathways in **plants**
INVENTOR(S): **Barbas, Carlos F.**; **Stege, Justin T.**; **Guan, Xueni**; Dalmia, Bipin
PATENT ASSIGNEE(S): USA
SOURCE: U.S. Pat. Appl. Publ., 84 pp., Cont.-in-part of U.S. Ser. No. 620,897.
CODEN: USXXCO
DOCUMENT TYPE: Patent
LANGUAGE: English
FAMILY ACC. NUM. COUNT: 2
PATENT INFORMATION:

PATENT NO.	KIND	DATE	APPLICATION NO.		DATE
US 2003037355	A1	20030220	US 2001-765555		20010119
PRIORITY APPLN. INFO.:			US 2000-177468P	P	20000121
			US 2000-620897	A2	20000721

AB The invention relates to the field of **plant** and agricultural technol. More specifically, the invention relates to the construction of **zinc finger proteins** and fusions of said proteins and their use to regulate gene expression and metabolic pathways in **plants**. **Plant** genes AP3 and MIPS were examd. for suitable zinc finger binding sites. The novel engineered **zinc finger proteins** used in the present methods are **ZFPm1**, **ZFPm2**, **ZFPm3**, **ZFPm4** and **ZFPAp3**. These proteins can be used alone or fused to an effector domain. The present methods can be used to modulate gene expression in monocot or dicot **plant** cells.

L22 ANSWER 2 OF 13 HCAPLUS COPYRIGHT 2003 ACS

ACCESSION NUMBER: 2003:229501 HCAPLUS
TITLE: Zinc fingers and a green thumb: manipulating

gene expression in plants
AUTHOR(S): Segal, David J.; **Stege, Justin T.**; **Barbas, Carlos F.**
CORPORATE SOURCE: The Skaggs Institute for Chemical Biology and the Department of Molecular Biology, The Scripps Research Institute, La Jolla, CA, 92037, USA
SOURCE: Current Opinion in Plant Biology (2003), 6(2), 163-168
CODEN: COPBFZ; ISSN: 1369-5266
PUBLISHER: Elsevier Science Ltd.
DOCUMENT TYPE: Journal
LANGUAGE: English
AB Artificial transcription factors can be rapidly constructed from predefined zinc-finger modules to regulate virtually any gene. Stable, heritable up- and downregulation of endogenous genes has been demonstrated in transgenic plants. These advances promise new approaches for creating functional knockouts and conditional overexpression, and for other gene discovery and manipulation applications in plants.

L22 ANSWER 3 OF 13 HCAPLUS COPYRIGHT 2003 ACS DUPLICATE 1
ACCESSION NUMBER: 2002:795317 HCAPLUS
DOCUMENT NUMBER: 138:84370
TITLE: Heritable endogenous gene regulation in **plants** with designed polydactyl zinc finger transcription factors
AUTHOR(S): **Guan, Xuen; Stege, Justin;** Kim, Myoung; Dahmani, Zina; Fan, Nancy; Heifetz, Peter; **Barbas, Carlos F., III;** Briggs, Steven P.
CORPORATE SOURCE: Torrey Mesa Research Institute, San Diego, CA, 92121, USA
SOURCE: Proceedings of the National Academy of Sciences of the United States of America (2002), 99(20), 13296-13301
CODEN: PNASA6; ISSN: 0027-8424
PUBLISHER: National Academy of Sciences
DOCUMENT TYPE: Journal
LANGUAGE: English
AB Zinc finger transcription factors (TFsZF) were designed and applied to transgene and endogenous gene regulation in stably transformed **plants**. The target of the TFsZF is the Arabidopsis gene APETALA3 (AP3), which encodes a transcription factor that dets. floral organ identity. A **zinc finger protein** (**ZFP**) was designed to specifically bind to a region upstream of AP3. AP3 transcription was induced by transformation of leaf protoplasts with a transformation vector that expressed a TFZF consisting of the **ZFP** fused to the tetrameric repeat of herpes simplex VP16's minimal activation domain. Histochem. staining of .beta.-glucuronidase (GUS) activity in transgenic AP3::GUS reporter **plants** expressing GUS under control of the AP3 promoter was increased dramatically in petals when the AP3-specific TFZF activator was cointroduced. TFZF-amplified GUS expression signals were also evident in sepal tissues of these double-transgenic **plants**. Floral phenotype changes indicative of endogenous AP3 factor coactivation were also obsd. The same AP3-specific **ZFPAP3** was also fused to a human transcriptional repression domain, the mSIN3

interaction domain, and introduced into either AP3::GUS-expressing **plants** or wild-type Arabidopsis **plants**. Dramatic repression of endogenous AP3 expression in floral tissue resulted when a constitutive promoter was used to drive the expression of this TFZF. These **plants** were also sterile. When a floral tissue-specific promoter from APETALA1 (AP1) gene was used, floral phenotype changes were also obsd., but in contrast the **plants** were fertile. Our results demonstrate that artificial transcriptional factors based on synthetic **zinc finger proteins** are capable of stable and specific regulation of endogenous genes through multiple generations in multicellular organisms.

REFERENCE COUNT: 50 THERE ARE 50 CITED REFERENCES AVAILABLE FOR THIS RECORD. ALL CITATIONS AVAILABLE IN THE RE FORMAT

L22 ANSWER 4 OF 13 HCAPLUS COPYRIGHT 2003 ACS DUPLICATE 2

ACCESSION NUMBER: 2002:795316 HCAPLUS
DOCUMENT NUMBER: 138:34056
TITLE: Regulation of transgene expression in **plants** with polydactyl zinc finger transcription factors
AUTHOR(S): Ordiz, M. Isabel; **Barbas, Carlos F., III**; Beachy, Roger N.
CORPORATE SOURCE: Donald Danforth Plant Science Center, St. Louis, MO, 63132, USA
SOURCE: Proceedings of the National Academy of Sciences of the United States of America (2002), 99(20), 13290-13295
CODEN: PNASA6; ISSN: 0027-8424
PUBLISHER: National Academy of Sciences
DOCUMENT TYPE: Journal
LANGUAGE: English

AB Designer zinc finger transcription factors (TFsZF) have been developed to control the expression of transgenes and endogenous genes in mammalian cells. Application of TFsZF technol. in **plants** would enable a wide range of both basic and applied studies. In this paper, we report the use of TFsZF to target a defined 18-bp DNA sequence to control gene expression in **plant** cells and in transgenic **plants**. A .beta.-glucuronidase reporter gene was activated by using the designed six-**zinc finger protein** 2C7 expressed as a fusion with the herpes simplex virus VP16 transcription factor activation domain. Reporter gene expression was activated 5- to 30-fold by using TFsZF in BY-2 protoplasts, whereas expression was increased as much as 450 times in transgenic **tobacco plants**. Use of a phloem-specific promoter to drive expression of the TFsZF resulted in activation of the reporter gene in vascular tissues. Transgenic **tobacco plants** that produce 2C7 transcription factors were phenotypically normal through two generations, suggesting that the factors exerted no adverse effects. This study demonstrates the utility of zinc finger technol. in **plants**, setting the stage for its application in basic and applied agricultural biotechnol.

REFERENCE COUNT: 25 THERE ARE 25 CITED REFERENCES AVAILABLE FOR THIS RECORD. ALL CITATIONS AVAILABLE IN THE RE FORMAT

L22 ANSWER 5 OF 13 HCAPLUS COPYRIGHT 2003 ACS DUPLICATE 3
ACCESSION NUMBER: 2003:81400 HCAPLUS
TITLE: Controlling gene expression in **plants** using synthetic zinc finger transcription factors
AUTHOR(S): **Stege, Justin T.; Guan, Xuen**; Ho, Thao; Beachy, Roger N.; **Barbas, Carlos F., III**
CORPORATE SOURCE: Department of Molecular Biology and The Skaggs Institute for Chemical Biology, Scripps Research Institute, La Jolla, CA, 92037, USA
SOURCE: Plant Journal (2002), 32(6), 1077-1086
CODEN: PLJUED; ISSN: 0960-7412
PUBLISHER: Blackwell Science Ltd.
DOCUMENT TYPE: Journal
LANGUAGE: English

AB Synthetic **zinc finger proteins** can be fused to transcriptional regulatory domains to create artificial transcription factors that modulate the expression of a specific target gene. Recent studies have demonstrated that synthetic zinc finger domains can be constructed to bind DNA sequences with a high degree of specificity. To devise a general strategy for controlling **plant** gene expression with artificial transcription factors, a rapid transient assay was developed to test the regulatory activity of synthetic zinc finger transcription factors (effectors) on target plasmids (reporters) in **plant** cells. Effective activation was demonstrated with **zinc finger proteins** fused to a deriv. of the VP16 activation domain. The mSin3 interaction domain (SID) of the human MAD1 protein provided moderate repression of target reporters. Unlike many naturally occurring transcription factors, these synthetic effectors exhibit a strong dependence on binding site position. Reporter genes that are stably integrated into **plant** cells responded similarly to transiently transfected reporter plasmids, verifying that this assay accurately reflects the behavior of these transcription factors on an endogenous target within the context of chromosomal DNA. These results provide evidence that synthetic **zinc finger proteins** can be used to manipulate the expression of endogenous genes in **plants**.

REFERENCE COUNT: 39 THERE ARE 39 CITED REFERENCES AVAILABLE FOR THIS RECORD. ALL CITATIONS AVAILABLE IN THE RE FORMAT

L22 ANSWER 6 OF 13 HCAPLUS COPYRIGHT 2003 ACS DUPLICATE 4
ACCESSION NUMBER: 2002:123927 HCAPLUS
DOCUMENT NUMBER: 137:42141
TITLE: Engineering polydactyl zinc-finger transcription factors
AUTHOR(S): Beerli, Roger R.; **Barbas, Carlos F., III**
CORPORATE SOURCE: The Skaggs Institute for Chem. Biology and Dep. of Molecular Biology, The Scripps Research Institute, La Jolla, CA, 92037, USA
SOURCE: Nature Biotechnology (2002), 20(2), 135-141
CODEN: NABIF9; ISSN: 1087-0156
PUBLISHER: Nature America Inc.
DOCUMENT TYPE: Journal; General Review
LANGUAGE: English

AB A review. The availability of rapid and robust methods for controlling gene function is of prime importance not only for assigning functions to newly discovered genes, but also for therapeutic intervention. Traditionally, gene function has been probed by often-laborious methods that either increase the level of a gene product or decrease it. Advances now make it possible to rapidly produce **zinc-finger proteins** capable of recognizing virtually any 18 bp stretch of DNA - a sequence long enough to specify a unique address in any genome. The attachment of functional domains also allows the design of tailor-made transcription factors for specific genes. Recent studies demonstrate that artificial transcription factors are capable of controlling the expression of endogenous genes in their native chromosomal context with a high degree of specificity in both animals and **plants**. Dominant regulatory control of expression of any endogenous gene can be achieved rapidly and can be also placed under chem. control. A wide range of potential applications is now within reach.

REFERENCE COUNT: 60 THERE ARE 60 CITED REFERENCES AVAILABLE FOR THIS RECORD. ALL CITATIONS AVAILABLE IN THE RE FORMAT

L22 ANSWER 7 OF 13 HCAPLUS COPYRIGHT 2003 ACS DUPLICATE 5
ACCESSION NUMBER: 2001:545414 HCAPLUS
DOCUMENT NUMBER: 135:133107
TITLE: Usage of **zinc finger protein** to regulate gene expression and metabolic pathways in **plants** and creation of five **zinc finger proteins**
INVENTOR(S): **Barbas, Carlos F., III; Stege, Justin T.; Guan, Xue Ni**; Dalmia, Bipin
PATENT ASSIGNEE(S): Scripps Research Institute, USA
SOURCE: PCT Int. Appl., 156 pp.
CODEN: PIXXD2
DOCUMENT TYPE: Patent
LANGUAGE: English
FAMILY ACC. NUM. COUNT: 2
PATENT INFORMATION:

PATENT NO.	KIND	DATE	APPLICATION NO.	DATE
WO 2001052620	A2	20010726	WO 2001-US1817	20010119
WO 2001052620	A3	20020207		
W: AE, AG, AL, AM, AT, AU, AZ, BA, BB, BG, BR, BY, CA, CH, CN, CR, CU, CZ, DE, DK, DM, DZ, EE, ES, FI, GB, GD, GE, GH, GM, HU, ID, IL, IN, IS, JP, KE, KG, KP, KR, KZ, LC, LK, LR, LS, LT, LU, LV, MA, MD, MG, MK, MN, MW, MX, NO, RU, TJ, TM				
RW: GH, GM, KE, LS, MW, MZ, SD, SL, UG, ZW, AT, BE, CH, CY, DE, DK, ES, FI, FR, GB, GR, IE, IT, LU, MC, NL, PT, SE, TR, BF, BJ, CF, CG, CI, CM, GA, GN, GW, ML, MR, NE, SN, TD, TG				
AU 2001029641	A5	20010731	AU 2001-29641	20010119
EP 1276869	A2	20030122	EP 2001-942508	20010119
R: AT, BE, CH, DE, DK, ES, FR, GB, GR, IT, LI, LU, NL, SE, MC, PT, IE, SI, LT, LV, FI, RO, MK, CY, AL, TR				

PRIORITY APPLN. INFO.: US 2000-177468P P 20000121
US 2000-620897 A 20000721

WO 2001-US1817 W 20010119

AB The invention relates to the field of **plant** and agricultural technol. More specifically, the invention relates to the use of **zinc finger proteins** and fusions of said proteins to regulate gene expression and metabolic pathways in **plants**. The genes, AP3 and MIPS, were examd. for suitable zinc finger binding sites. Five new **zinc finger proteins, ZFPAp3, ZFPm1**, **ZFPm2, ZFPm3** and **ZFPm4**, were constructed from human **zinc finger protein** Sp1C, expressed in E. coli and purified. DNA binding specificity of **ZFPAp3, ZFPm1, ZFPm2, ZFPm3** and **ZFPm4** was characterized.

L22 ANSWER 8 OF 13 HCAPLUS COPYRIGHT 2003 ACS
ACCESSION NUMBER: 2001:408061 HCAPLUS
DOCUMENT NUMBER: 135:30537
TITLE: Design, construction and of **zinc finger protein** derivatives and their use in the modulation of gene expression
INVENTOR(S): **Barbas, Carlos F., III**; Gottesfeld, Joel M.; Wright, Peter E.
PATENT ASSIGNEE(S): The Scripps Research Institute, USA
SOURCE: U.S., 67 pp., Cont.-in-part of U.S. Ser. No. 312,604, abandoned.
CODEN: USXXAM
DOCUMENT TYPE: Patent
LANGUAGE: English
FAMILY ACC. NUM. COUNT: 2
PATENT INFORMATION:

PATENT NO.	KIND	DATE	APPLICATION NO.	DATE
US 6242568	B1	20010605	US 1996-676318	19961230
WO 9519431	A1	19950720	WO 1995-US829	19950118

W: AU, CA, FI, JP, NO, US
RW: AT, BE, CH, DE, DK, ES, FR, GB, GR, IE, IT, LU, MC, NL, PT, SE

PRIORITY APPLN. INFO.: US 1994-183119 B2 19940118
US 1994-312604 B2 19940928
WO 1995-US829 W 19950118

AB The present invention provides zinc finger nucleotide binding protein variants that have at least two zinc finger modules that bind to a target cellular nucleotide sequence and modulate the transcriptional function of the cellular nucleotide sequence. Also provided are methods of use of such zinc finger nucleotide binding protein variants and methods for isolating the same using expression libraries encoding the protein variants contg. randomized substitutions of amino acids. Exemplary zinc finger nucleotide binding protein variants of the invention include two cysteines and two histidines whereby both cysteines are amino proximal to both histidines. Design and construction of variants of the **zinc finger protein** Zif/268 are disclosed. Construction of multifinger proteins utilizing repeats of the first finger of Zif/268 is described.
REFERENCE COUNT: 28 THERE ARE 28 CITED REFERENCES AVAILABLE FOR THIS RECORD. ALL CITATIONS AVAILABLE

IN THE RE FORMAT

L22 ANSWER 9 OF 13 WPIDS (C) 2003 THOMSON DERWENT
ACCESSION NUMBER: 2001-308618 [32] WPIDS
DOC. NO. CPI: C2001-095392
TITLE: New fusion protein containing nucleotide-binding and ligand-binding domains, useful e.g. in gene therapy of cancer, provides ligand-activated control of gene expression.
DERWENT CLASS: B04 D16
INVENTOR(S): **BARBAS, C F**; BEERLI, R; KADAN, M
PATENT ASSIGNEE(S): (NOVS) NOVARTIS AG; (SCRI) SCRIPPS RES INST
COUNTRY COUNT: 94
PATENT INFORMATION:

PATENT NO	KIND	DATE	WEEK	LA	PG
WO 2001030843	A1	20010503	(200132)*	EN	217

RW: AT BE CH CY DE DK EA ES FI FR GB GH GM GR IE IT KE LS LU MC MW MZ NL OA PT SD SE SL SZ TZ UG ZW
W: AE AG AL AM AT AU AZ BA BB BG BR BY BZ CA CH CN CR CU CZ DE DK DM DZ EE ES FI GB GD GE GH GM HR HU ID IL IN IS JP KE KG KP KR KZ LC LK LR LS LT LU LV MA MD MG MK MN MW MX MZ NO NZ PL PT RO RU SD SE SG SI SK SL TJ TM TR TT TZ UA UG UZ VN YU ZA ZW

AU 2001011438	A	20010508	(200149)	
EP 1226168	A1	20020731	(200257)	EN

R: AL AT BE CH CY DE DK ES FI FR GB GR IE IT LI LT LU LV MC MK NL PT RO SE SI

APPLICATION DETAILS:

PATENT NO	KIND	APPLICATION	DATE
WO 2001030843	A1	WO 2000-EP10430	20001023
AU 2001011438	A	AU 2001-11438	20001023
EP 1226168	A1	EP 2000-972849	20001023
		WO 2000-EP10430	20001023

FILING DETAILS:

PATENT NO	KIND		PATENT NO
AU 2001011438	A	Based on	WO 200130843
EP 1226168	A1	Based on	WO 200130843

PRIORITY APPLN. INFO: US 2000-586625 20000602; US 1999-433042 19991025
AN 2001-308618 [32] WPIDS
AB WO 200130843 A UPAB: 20010611
NOVELTY - Fusion protein (I) comprising a nucleotide-binding domain (NBD) linked to a ligand-binding domain (LBD) of an intracellular receptor (ICR). NBD is a polydactyl **zinc finger protein**, or a modular part of it, that interacts specifically with a contiguous sequence of at least 3 nucleotides (nt), and (I) functions as a ligand-activated transcriptional regulator.
DETAILED DESCRIPTION - INDEPENDENT CLAIMS are also included

fort he following:
(a) nucleic acid (II) that encodes (I);
(b) vector containing (II);
(c) cell containing the vector of (b);
(d) combination of (I) or (II) with a regulatable expression cassette containing at lest one response element recognized by NBD;
(e) composition for regulating gene expression comprising (I) or (II) plus an excipient;
(f) regulating gene expression in a cell by introducing (I) or (II) then treating the cell with a ligand that interacts with LBD; and
(g) non-viral delivery system comprising (I) or (II).
ACTIVITY - Anticancer; Antiproliferative.
MECHANISM OF ACTION - Ligand-activated regulation of transcription.
USE - (I), or the nucleic acid (II) that encodes it, is used to regulate gene expression, particularly in gene therapy, especially of malignant or non-malignant proliferative disease (cancer, psoriasis, Behcet syndrome etc.), e.g. where induced by viruses in humans or **plants**, also genetic and/or acquired diseases.
ADVANTAGE - (I) can be designed to target any selected gene (endogenous or exogenous), and can be made to have different selectivity or specificity for endogenous or exogenous ligands.
Dwg.0/27

L22 ANSWER 10 OF 13 BIOSIS COPYRIGHT 2003 BIOLOGICAL ABSTRACTS INC.
ACCESSION NUMBER: 2001:249053 BIOSIS
DOCUMENT NUMBER: PREV200100249053
TITLE: **Zinc finger protein** derivatives and methods therefor.
AUTHOR(S): **Barbas, Carlos F.**; Gottesfeld, Joel M. (1); Wright, Peter E.
CORPORATE SOURCE: (1) Del Mar, CA USA
ASSIGNEE: The Scripps Research Institute
PATENT INFORMATION: US 6140466 October 31, 2000
SOURCE: Official Gazette of the United States Patent and Trademark Office Patents, (Oct. 31, 2000) Vol. 1239, No. 5, pp. No Pagination. e-file.
ISSN: 0098-1133.
DOCUMENT TYPE: Patent
LANGUAGE: English
AB **Zinc finger proteins** of the Cys2 His2 type represent a class of malleable DNA binding proteins which may be selected to bind diverse sequences. Typically, **zinc finger proteins** containing three zinc finger domains, like the murine transcription factor Zif268 and the human transcription factor Sp1, bind nine contiguous base pairs (bp). To create a class of proteins which would be generally applicable to target unique sites within complex genomes, the present invention provides a polypeptide linker that fuses two three-finger proteins. Two six-fingered proteins were created and demonstrated to bind 18 contiguous bp of DNA in a sequence specific fashion. Expression of these proteins as fusions to activation or repression domains allows transcription to be specifically up or down modulated within cells. Polydactyl **zinc finger proteins** are broadly applicable as genome-specific transcriptional switches in gene therapy strategies and the development of novel transgenic **plants** and animals. Such proteins are useful for inhibiting,

activating or enhancing gene expression from a zinc finger-nucleotide binding motif containing promoter or other transcriptional control element, as well as a structural gene or RNA sequence.

L22 ANSWER 11 OF 13 SCISEARCH COPYRIGHT 2003 ISI (R)
ACCESSION NUMBER: 1999:237427 SCISEARCH
THE GENUINE ARTICLE: 177RH
TITLE: Toward controlling gene expression at will: Selection and design of zinc finger domains recognizing each of the 5'-GNN-3' DNA target sequences
AUTHOR: Segal D J; Dreier B; Beerli R R; **Barbas C F (Reprint)**
CORPORATE SOURCE: SCRIPPS RES INST, SKAGGS INST CHEM BIOL, BCC-515, 10550 N TORREY PINES RD, LA JOLLA, CA 92037 (Reprint); SCRIPPS RES INST, SKAGGS INST CHEM BIOL, LA JOLLA, CA 92037; SCRIPPS RES INST, DEPT BIOL MOL, LA JOLLA, CA 92037
COUNTRY OF AUTHOR: USA
SOURCE: PROCEEDINGS OF THE NATIONAL ACADEMY OF SCIENCES OF THE UNITED STATES OF AMERICA, (16 MAR 1999) Vol. 96, No. 6, pp. 2758-2763.
Publisher: NATL ACAD SCIENCES, 2101 CONSTITUTION AVE NW, WASHINGTON, DC 20418.
ISSN: 0027-8424.
DOCUMENT TYPE: Article; Journal
FILE SEGMENT: LIFE
LANGUAGE: English
REFERENCE COUNT: 35

ABSTRACT IS AVAILABLE IN THE ALL AND IALL FORMATS

AB We have taken a comprehensive approach to the generation of novel DNA binding zinc finger domains of defined specificity, Herein **rye** describe the generation and characterization of a family of zinc finger domains developed for the recognition of each of the 16 possible 3-bp DNA binding sites having the sequence 5'-GNN-3'. Phage display libraries of **zinc finger proteins** were created and selected under conditions that favor enrichment of sequence-specific proteins. Zinc finger domains recognizing a number of sequences required refinement by site-directed mutagenesis that was guided by both phage selection data and structural information. In many cases, residues not expected to make base-specific contacts had effects on specificity. A number of these domains demonstrate exquisite specificity and discriminate between sequences that differ by a single base with >100-fold loss in affinity, We conclude that the three helical positions -1, 3, and 6 of a zinc finger domain are insufficient to allow. for the fine specificity of the DNA binding domain to be predicted. These domains are functionally modular and may be recombined with one another to create polydactyl proteins capable of binding 18-bp sequences with subnanomolar affinity The family of zinc finger domains described here is sufficient for the construction of 17 million novel proteins that bind the 5'-(GNN)(6)-3' family of DNA. sequences. These materials and methods should allow for the rapid construction of novel gene sa itches and provide the basis for a universal system for gene control.

L22 ANSWER 12 OF 13 HCAPLUS COPYRIGHT 2003 ACS

ACCESSION NUMBER: 1998:795122 HCAPLUS
DOCUMENT NUMBER: 130:33969
TITLE: Design and construction of **zinc finger protein** derivatives
INVENTOR(S): **Barbas, Carlos F., III**; Gottesfeld, Joel M.; Wright, Peter E.
PATENT ASSIGNEE(S): The Scripps Research Institute, USA
SOURCE: PCT Int. Appl., 159 pp.
CODEN: PIXXD2
DOCUMENT TYPE: Patent
LANGUAGE: English
FAMILY ACC. NUM. COUNT: 1
PATENT INFORMATION:

PATENT NO.	KIND	DATE	APPLICATION NO.	DATE
WO 9854311	A1	19981203	WO 1998-US10801	19980527
W: AL, AM, AT, AU, AZ, BA, BB, BG, BR, BY, CA, CH, CN, CU, CZ, DE, DK, EE, ES, FI, GB, GE, GH, GM, GW, HU, ID, IL, IS, JP, KE, KG, KP, KR, KZ, LC, LK, LR, LS, LT, LU, LV, MD, MG, MK, MN, MW, MX, NO, NZ, PL, PT, RO, RU, SD, SE, SG, SI, SK, SL, TJ, TM, TR, TT, UA, UG, US, UZ, VN, YU, ZW, AM, AZ, BY, KG, KZ, MD, RU, TJ, TM				
RW: GH, GM, KE, LS, MW, SD, SZ, UG, ZW, AT, BE, CH, CY, DE, DK, ES, FI, FR, GB, GR, IE, IT, LU, MC, NL, PT, SE, BF, BJ, CF, CG, CI, CM, GA, GN, ML, MR, NE, SN, TD, TG				
US 6140466	A	20001031	US 1997-863813	19970527
AU 9878003	A1	19981230	AU 1998-78003	19980527
EP 988377	A1	20000329	EP 1998-926088	19980527
R: AT, BE, CH, DE, DK, ES, FR, GB, GR, IT, LI, LU, NL, SE, MC, PT, IE, FI				
JP 2002502249	T2	20020122	JP 1999-500870	19980527

PRIORITY APPLN. INFO.: US 1997-863813 A1 19970527
WO 1998-US10801 W 19980527

AB **Zinc finger proteins** of the Cys2His2 type represent a class of malleable DNA-binding proteins which may be selected to bind diverse sequences. Typically, **zinc finger proteins** contg. 3 zinc finger domains, like the murine transcription factor Zif268 and the human transcription factor Sp1, bind 9 contiguous base pairs (bp). To create a class of proteins which would be generally applicable to target unique sites within complex genomes, the present invention provides a polypeptide linker (Thr-Gly-Glu-Lys-Pro) that fuses two 3-finger proteins. Two 6-fingered proteins were created and demonstrated to bind 18 contiguous bp of DNA in a sequence-specific fashion. Expression of these proteins as fusions to activation or repression domains (e.g., with Jun/Fos leucine zipper domains, the Kruppel-assocd. box A domain, or the transcriptional activation domain of herpes simplex virus VP16 protein) allows transcription to be specifically up- or down-modulated within cells. Polydactyl **zinc finger proteins** are broadly applicable as genome-specific transcriptional switches in gene therapy strategies and the development of novel transgenic **plants** and animals. Such proteins are useful for inhibiting, activating or enhancing gene expression from a zinc finger-nucleotide binding motif contg. promoter or other transcriptional control element, as well as a structural gene or RNA sequence.

REFERENCE COUNT: 18 THERE ARE 18 CITED REFERENCES AVAILABLE

FOR THIS RECORD. ALL CITATIONS AVAILABLE IN THE RE FORMAT

L22 ANSWER 13 OF 13 HCAPLUS COPYRIGHT 2003 ACS DUPLICATE 6
ACCESSION NUMBER: 1997:356607 HCAPLUS
DOCUMENT NUMBER: 127:62222
TITLE: Design of polydactyl **zinc-finger proteins** for unique addressing within complex genomes
AUTHOR(S): Liu, Qiang; Segal, David J.; Ghiara, Jayant B.; **Barbas, Carlos F., III**
CORPORATE SOURCE: Skaggs Inst. Chem. Biol. and Dep. Molecular Biol., Scripps Res. Inst., La Jolla, CA, 92037, USA
SOURCE: Proceedings of the National Academy of Sciences of the United States of America (1997), 94(11), 5525-5530
CODEN: PNASA6; ISSN: 0027-8424
PUBLISHER: National Academy of Sciences
DOCUMENT TYPE: Journal
LANGUAGE: English

AB **Zinc-finger proteins** of the Cys2-His2 type represent a class of malleable DNA-binding proteins that may be selected to bind diverse sequences. Typically, **zinc-finger proteins** contg. three zinc-finger domains, like the murine transcription factor Zif268 and the human transcription factor Sp1, bind nine contiguous base pairs. To create a class of proteins that would be generally applicable to target unique sites within complex genomes, the authors have utilized structure-based modeling to design a polypeptide linker that fuses two three-finger proteins. Two six-fingered proteins were created and demonstrated to bind 18 contiguous bp of DNA in a sequence-specific fashion. Expression of these proteins as fusions to activation or repression domains allows transcription to be specifically up- or down-modulated within human cells. Polydactyl **zinc-finger proteins** should be broadly applicable as genome-specific transcriptional switches in gene therapy strategies and the development of novel transgenic **plants** and animals.

FILE 'HOME' ENTERED AT 10:59:21 ON 27 MAR 2003